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THE PHARMACOLOGY OF CURARE AND CURARISING SUBSTANCES

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Historical

Eighty years have now elapsed since Crum Brown and Fraser[1,2], in a paper still worth close study, laid the foundations of our knowledge of the relation between the chemical structure and the pharmacological action of quaternary salts. Even at that early date, they referred to the therapeutic possibilities of the new compounds they had described, and to their advantages over curare in being "readily obtained in a state of perfect purity, and, therefore, of constant strength." It is only in recent years, however, that either a natural alkaloid or a synthetic curarising substance has emerged in good supply and of the required properties. This rapid progress has been greatly stimulated by King's determination of the structure of *d*-tubocurarine chloride[3,4,5] by the success of Bennett[6] in softening therapeutic convulsions with a curare extract prepared by MacIntyre and standardised by Holaday's rabbit head-drop method; by Griffith and Johnson's pioneer demonstrations of the usefulness of curare in anæsthesia[7]; by Wintersteiner and Dutcher's isolation[8] of a potent curarising extract (intocostrin) rich in *d*-tubocurarine chloride, from a known botanical species (*Chondrodendron tomentosum*); and by the discovery of Bovet and his colleagues of synthetic compounds with a potency comparable to that of the natural alkaloids[9,10,11,12,13]. As a result of this and other work, the clinician now commands a choice of reliable and well-studied compounds, and the pharmacologist has been enriched by many stimulating (and often bewildering) additions to his knowledge.

Recent Literature

Four important reviews should be consulted for detailed references: Ing has reviewed the relation of the chemical structure of onium salts to their pharmacological action in an authoritative article[14]. MacIntyres' "Curare"[15] is a very valuable source-book, particularly on the history of the subject: Bovet and Bovet-Nitti[12] have reviewed recent work, particularly their own studies of "curares de synthese": and Craig[16] has compiled and discussed a very extensive list of compounds tested up till 1947 for curarising action. There are many reports of use in medicine, surgery and anæsthesia, for which reference should be made to the clinical journals. A recent discussion by Kuffler, Acheson, Welsh and Harvey[17] of theories of neuromuscular transmission also deserves attention.

The present article is not intended to be a comprehensive survey of the subject, but attempts to review important features of the work of

recent years in the light of previous knowledge, and to indicate some of the problems remaining unsolved.

Modes of "Curarisation"

The term "curarisation" is no longer restricted in current use to that form of neuromuscular block caused by curare, but is sometimes used where similar effects are produced by other drugs, even where no more than muscular relaxation is meant (e.g., the action of myanesin). This implies, of course, a considerable widening of the meaning of the word, and a corresponding risk of confusion. But such extension of meaning is not altogether inappropriate; for block at the neuromuscular junction can be regarded as a special case of synaptic block, which may also occur at the ganglionic or central nervous synapse. Dale[18] and Feldberg[19] have discussed the evidence that the transmissions at each of these synapses possess features in common, with particular reference to the possibility of a common mechanism of chemical transmission. It should be noted that if this evidence is accepted, the failure of curarising substances to exert similar actions at each type of synapse is a fact of the first importance. For the time being, however, the interests of clarity are best served by simply describing, with particular reference to the neuromuscular junction, the different kinds of synaptic block that can be referred to as curarisation. The opportunity will be taken, in doing this, of mentioning some of the recent additions to knowledge in this field.

1. The Neuromuscular Junction. (a) *The action of curare alkaloids.* Classical experiments have established that after paralysis of a muscle by curare to excitation through its nerve, conduction in the nerve trunk is unaltered and the muscle can still give a propagated contraction in response to direct electrical stimulation. The site of the paralysis is thus localised to the nerve terminals and motor end-plate. Dale and his colleagues have further shown that the terminal nerve endings of the curarised muscle still liberate acetylcholine, and that arterially injected acetylcholine is antagonised as much as, or more than, the effect of a nerve volley. Apart from the relevance of these facts to the mechanism of neuromuscular transmission, they are also important criteria of curare-like action. To them may be added the inhibition by curare of the contracture of frog's rectus due to acetylcholine; the antagonism of anticholinesterases to the actions of curare on the neuromuscular junction; the failure of the partially curarised muscle to sustain a tetanus; and the revealing, under suitable conditions, of a transient potential at the end-plate excited by nerve stimulation, which has been closely studied and termed the "end-plate potential" by Eccles and his colleagues. These, among other characteristics, constitute a highly specific picture.

The normal conception of neuromuscular transmission at present is as follows: the wave of excitation reaches the nerve terminals, and there causes the discharge of acetylcholine in close relation to the motor end-plate. This discharge depolarises the end-plate (giving rise to the end-plate potential) and the depolarisation of the end-plate in turn excites the

muscle fibre. If this conception is accepted, then the characteristic actions of curare lend themselves very readily to the belief that curare acts by raising the threshold of the motor end-plate to excitation by acetylcholine, and that it exerts this action by competing for the acetylcholine receptor sites on the end-plate. But although this view of its action is both plausible and widely held, it cannot be said to be established beyond all question, for there are many phenomena of curarisation that still remain unexplained. It certainly provides, however, a most useful working hypothesis.

It is unfortunate that the specific tests mentioned are rarely used to verify that a substance is "curare-like"; proof of excitability of nerve and muscle is commonly omitted, and investigation of effect on acetylcholine release is a rarity. One test sometimes used, that of antagonism by anticholinesterases, is, by itself, useless, since these substances may greatly increase the tension of the twitch of the normal uncurarised muscle.

(b) *The action of* bis*trimethylammonium decane diiodide* (*C*10). This compound, which will be further discussed below, is curare-like in four respects: during its action, conduction in nerve is unaltered, direct excitability of muscle is retained, injected acetylcholine is rendered ineffective, and release of acetylcholine by motor nerve stimulation is not prevented. But there are also important differences; C10 itself elicits a contraction of frog's rectus; it is not antagonised by anticholinesterases, although C5 (the pentane homologue) is an effective antagonist; its activity varies very greatly with species of animal used for test; and it produces a depolarisation of the muscle membrane[20]. None of these effects is shown by curare. The mode of action of C10 is still uncertain, but these and other differences from curare are sufficiently great to make it necessary to distinguish the actions of the two drugs.

(c) *The action of anticholinesterases.* Eserine has long been known as a depressant of the muscular contraction caused by a tetanus of the motor nerve, although it usually augments the tension of single twitches. This depressant action exerted both by eserine and by other anticholinesterases, is due to the accumulation of paralysing concentrations of acetylcholine at the end-plate (Brown, Dale and Feldberg[21]). It is not possible, however, to exclude entirely some direct action by the anticholinesterase itself (*cf.* Riker and Wescoe[22]).

(d) *The action of substances depressing the release of acetylcholine.* Harvey[23] has presented evidence suggesting that some of the neuromuscular block caused by procaine is due to interference with release of acetylcholine by the nerve-ending. A similar block is caused by botulinus toxin (Burgen, Dickens and Zatman[24]) after which progressive failure of transmission occurs, although nerve and muscle remain excitable: acetylcholine injected is still effective, but release of acetylcholine is depressed. Brown and Harvey[25] and Brown and Vianna Dias[26] have reported that such a failure of acetylcholine release also results from calcium deficiency or from injection or perfusion with solutions rich in phosphate.

2. The Ganglionic Synapse. Depression of transmission at the synapse of the superior cervical ganglion by curare, without loss of excitability of preganglionic fibres or of ganglion cells, or abolition of the release of acetylcholine has been shown by Brown and Feldberg[27]. Brown and Feldberg[28] have also demonstrated block of transmission due to accumulation of acetylcholine in the presence of eserine. Harvey[23] found that procaine blocked transmission by preventing release of acetylcholine at preganglionic nerve terminals, and Harvey and MacIntosh[29] have shown that calcium lack in the perfusion fluid leads to the same result. Block by injection of large doses of potassium has also been described (Brown and Feldberg[30]).

The same types of "curarisation" may, therefore, be seen in the ganglion as at the neuromuscular junction. One of the most interesting developments recently, however, has been the observation that a "curarising" compound which is active at the neuromuscular junction may be relatively inactive on the ganglion and *vice versa*. It has, of course, been known for some time that tetraethylammonium iodide, although of neglible activity at the neuromuscular junction, is a powerful paralyser of ganglionic transmission (Burn and Dale[31], Acheson and Moe[32]). Depierre[33] studied this point on certain of Bovet's ethyl-choline ethers of phenol and polyphenols, using the contraction of the cat's nictitating membrane excited by stimulation of the cervical sympathetic. These compounds can be arranged in a series in which curarising activity increases as ganglionic activity diminishes (so that the ratio of curarising dose to ganglion-paralysing dose ranges from *c.* 30 to 0·02). A similar dissociation occurs in the *bis*trimethylammonium series: here C5 or C6 injected intravenously into the cat requires only 1·0 mg./kg. to affect the superior cervical ganglion, but 10 to 20 mg./kg. or more are required to depress neuromuscular conduction; C10, on the other hand, active at the junction in a dose of 30μg./kg. requires more than 3 mg./kg. to depress ganglionic transmission. The ratio just mentioned is thus more than 100 for C5 and less than 0·01 for C10. It is clear from these and similar results that activities in paralysing neuromuscular transmission and ganglionic transmission can be very widely dissociated.

3. The Central Nervous Synapse. No method yet exists adequate to determine whether a given depression of activity by the spinal cord or brain is due to paralysis of transmission at the synapse rather than to failure of conduction in neurone or axon. The action of a drug such as myanesin however, suggests some such action, since it can depress reflex activity without depressing conduction in a peripheral nerve. The point most relevant, at the moment, is the remarkable lack of central action of drugs such as *d*-tubocurarine chloride or C10. Some of this inactivity may be due to failure to pass through the capillaries of the central nervous system, which cations traverse but slowly (Krogh[34]). But even when curare is administered more directly, it appears most commonly to exert a central stimulant action. It is possible, therefore, that the central synapse is, like the neuromuscular and ganglionic synapses, relatively specific as regards the agents which block it; we have to contrast drugs

like curare (notably paralytic on the neuromuscular junction and ganglionic synapse, but centrally stimulant); tetraethylammonium iodide (almost inactive at the junction, highly active on the superior cervical ganglion, and of mixed action on the central nervous system) (Salama[35]); and myanesin (inactive on neuromuscular conduction but centrally depressant).

In referring, therefore, to "curarisation" in its extended sense, it is necessary to specify both the synapse at which paralysis of transmission of excitation occurs, and the mode of that paralysis. A tentative summary of the modes of paralysis of transmission due to various drugs described can be attempted. (a) Competition block (e.g., curare) in which the threshold of the end-plate to acetylcholine is raised; (b) depolarisation block (e.g. potassium chloride and possibly C10); (c) block by accumulation of acetylcholine (e.g. anticholinesterases); (d) block by transmitter failure, either by immobilisation of acetylcholine (e.g. procaine, calcium lack), or by deficiency of acetylcholine (e.g. botulinus toxin). This variety of modes of "curarisation" makes it essential to define clearly which is being used, even if the fundamental mechanism of the particular type of block is not fully understood. These distinctions become all the more necessary if any attempt is to be made to relate "curarising" potency to chemical structure. For the purpose of this review, the distinctions just made will not be pursued, and discussion will be confined to the pharmacology of compounds producing block of curare-like or C10-like character.

Natural Alkaloids

1. The Curare Group. The total number of such alkaloids is very large. For practical purposes three should be carefully distinguished: (a)

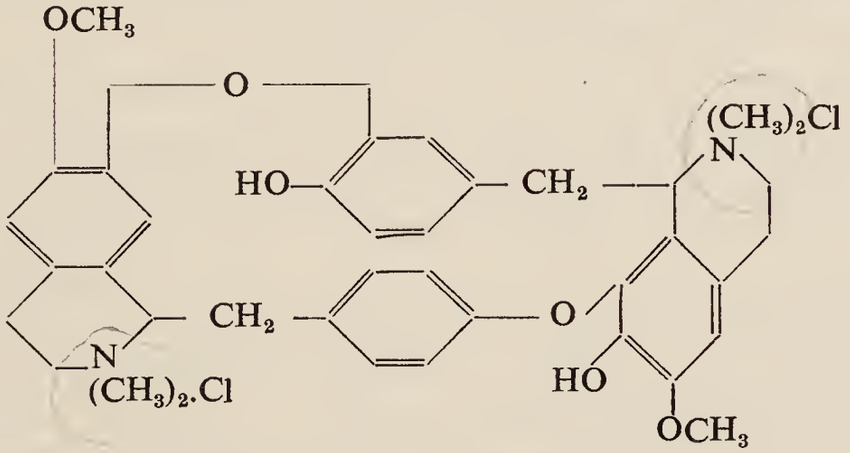


Fig. 1. Structure of *d*-Tubocurarine chloride.

curarine chloride; this is the material isolated by Boehm from calabash curare, much used in experimental work before the isolation of *d*-tubocurarine chloride: (b) *d*-tubocurarine chloride (*d*-T.C.), isolated by King[2] from tube curare, whose structure was finally determined by him[4]: (c) intocostrin, an extract prepared by Squibbs, rich in *d*-tubocurarine chloride; the intocostrin unit is equivalent to 1 mg. of a standard preparation from *Chondrodendron tomentosum,* and the activity of 6·5 units is equal to that of 1 mg. of *d*-tubocurarine chloride, on the rabbit head-drop test. "Curare" is used in this review as a generic term when the distinction between the above substances is not important.

The main features of the pharmacology of these substances at the neuromuscular junction have already been described; further details will be found in the references cited. But other properties of the compounds have recently come into prominence arising particularly with reference to the possibility of side-actions in clinical use.

(a) *Liberation of histamine.* The observation that curarine liberated histamine from muscle by Alam, Anrep, Barsoum, Talaat and Weininger[36] has been repeated by Gregory and Schild[37]; the latter showed that *d*-T.C. exerted this action both on the perfused tongue of the cat and on the rat's diaphragm. Grob, Lilienthal and Harvey[38] have extended these results to man, showing that *d*-T.C. injected into the brachial artery causes flushing, œdema, and itching of the arm, which is lessened by anti-histamines, and that injected intradermally it causes a typical " triple response." Striking confirmation of this work was provided by Landmesser[39], who showed that *d*-T.C. caused fall of blood pressure in spinal dogs and bronchoconstriction of guinea-pig's lung similar to that caused by peptone, and that these effects were prevented by antihistamines. Further, he observed that occasional dogs were refractory to the effects of *d*-T.C. (just as they are to peptone), and that sensitive animals could be made refractory to peptone by previous injection of adequate doses of *d*-T.C., or *vice-versa*. *d*-T.C. thus resembles peptone rather closely, and it is highly probable that, as with peptone, the liberation of heparin accompanies the liberation of histamine by the drug. As Landmesser points out, the practical application of these results is difficult, since, for instance, anæsthetics depress the release of histamine. But it is clearly desirable, at least, that compounds less active in this respect should be investigated.

(b) *Actions on the C.N.S.* That *d*-T.C. has some central stimulant property has already been mentioned. The experimental results in this field, however, are still somewhat confused. References to the stimulant action of curare when applied directly to the central nervous system may be found in the reviews mentioned. Salama[35] has recently verified these results, administering *d*-T.C. directly into the ventricles of cats. It is common to see a stage of excitement preceding curarisation with *d*-T.C., particularly with smaller animals. Everett[40] has described the convulsant action of *d*-T.C. given intracisternally to rabbits. On the other hand, it also has been claimed that curare given intravenously may have a narcotic or anæsthetic action. Whitacre and Fisher[41] report an illustrative case from their surgical experience. The report by Prescott, Organe and Rowbotham[42], however, and the careful and courageous experiment by Smith, Brown, Toman and Goodman[43] fail to substantiate this. In the latter paper, an account is given of the curarisation of a volunteer, under artificial respiration, so deeply that not even the most trivial muscular movement could be made; nevertheless, a full and intelligent narrative of his experience was furnished by the subject after recovery. Kellgren, McGowan and Wood[44] found no alteration in sensation after small doses of *d*-T.C. Paton and Zaimis[45], studying

the respiratory depression by *d*-T.C., found that the discharge down the phrenic nerve of the cat was not depressed by an intravenous dose of *d*-T.C. sufficient to abolish spontaneous respiration. It seems unlikely from this and other work, therefore, that the central actions of *d*-T.C. are important after intravenous doses, probably because (as mentioned above) it would not be expected to pass very readily through the capillaries of the central nervous system.

(c) *Action on autonomic ganglia.* Curare has long been known to depress ganglionic transmission, as has been already mentioned. Such action has, indeed, been suggested as the basis of an assay method, in which inhibition of the peristaltic reflex of isolated intestine is used to assay *d*-T.C. or kindred drugs (Feldberg and Lin[46]). Gross and Cullen[47] have shown that in the dog, curarising doses of intocostrin or *d*-T.C. cause inhibition of peristaltic activity by stomach and small intestine, with some loss of tone; fall of blood pressure was also sometimes observed. Heymans[48,49] found that rapid injections of intocostrin cause a fall of blood pressure and depression of the cardiovascular reflexes, but that with slow injection of the same dose these actions did not appear. Prostigmine did not abolish these effects.

(d) *Antagonism by certain dyes.* An old observation that certain dyestuffs antagonise curare has been reinvestigated by Kensler[50]. Congo red, chlorazol fast pink and Evans Blue are highly effective at both preventing and relieving paralysis of frogs by *d*-T.C. The action is due to the formation of a precipitable complex, in which form *d*-T.C. is not active. The phenomenon promises to be a useful tool in suitable circumstances.

(e) *Anticholinesterase action.* Some of the earlier preparations of intocostrin contained material with an appreciable power of inhibiting cholinesterase (Harris and Harris[51]). Pure *d*-T.C., however, has slight activity in this respect; the materials responsible were tertiary bases of negligible curarising activity. The finding is of interest, in view of the extent to which anticholinesterase action has been observed among synthetic compounds.

2. Dimethyl Ether Of *d*-Tubocurarine Chloride. It has been known for some time that methylation of *d*-T.C. increases its potency. Further studies (Collier, Paris and Woolf[52]) have shown that the dimethyl ether is about 10 times as active as *d*-T.C. in rabbits, and that it displays certain species differences in potency and duration of action. In the main, it is very similar to *d*-T.C., but considerably more active. Successful clinical trials have been reported by Stoelting, Graf and Viera[53].

3. The Erythrina Alkaloids. Exceptional interest attaches to these compounds, of which erythroidine (from the seeds of the legume *Erythrina Americana*) and β-dihydroerythroidine (obtained by hydrogenating erythroidine) are the most important. (Erythrina extracts are said to have been used in the treatment of convulsions as long ago as 1887.) They are at present unique in being highly active and yet possessing only trivalent nitrogen atoms; on converting the latter to quaternary nitrogen, the compounds diminish greatly in potency. So far as their

action at the neuromuscular junction is concerned, they resemble the curare alkaloids, having 1/5th or less the activity of *d*-tubocurarine chloride, and they are antagonised by anticholinesterases. In other respects, there are important differences; the erythroidines are active by mouth; they do not possess anti-esterase activity (Harris and Harris[51]); they do not share the ability of *d*-tubocurarine to liberate histamine (Landmesser[39]); they are not antagonised by congo red (Kensler[50]); they possess a feeble atropine-like action. Clinical trial has been reported (Harvey and Masland[54], Dripps and Sergent[55]); the most serious disadvantage appears to be a depression of the blood pressure with effective doses; respiratory depression is also common. It is to be hoped that other members of this series may be discovered which are free from these defects.

4. Other Natural Alkaloids And Their Derivatives. The comprehensive study of quinine methochloride by Harvey[56] requires mention; this is one of the few compounds investigated whose action on the release of acetylcholine has been tested; in paralysing doses, it fails to prevent such release in the superior cervical ganglion of the cat. It is curare-like in most respects, is about 1/40 times as active as *d*-T.C., and is active by mouth. Trials in man have been reported by Harvey and Masland[54]. A large number of other related alkaloids of the cinchona group have also been studied but are not promising clinically.

Among compounds related to *d*-T.C., the *iso*chondrodendrines (Marsh and Pelletier[57]) and the chondrodendrines (Marsh, Sleeth and Tucker[58]) have been investigated; in both groups variation of potency with species, and increase in potency by methylating free hydroxyl groups were found, analogous with the effect of methylating *d*-tubocurarine chloride. Another related compound, N-methyl oxyacanthine (Marsh, Herring and Sleeth[59]) is of interest, since in man it lessens salivary secretion, and has a weak atropine-like action (antagonising the depression of dog's blood pressure by acetylcholine).

The most potent known curarising substances are the toxiferines, isolated by Wieland from calabash curare; they are effective in doses of the order of 10μg./kg. in frogs and rabbits. So far as is known, however, they do not appear to be sufficiently free from side-actions of various kinds to be suitable for therapeutic use.

On the whole, therefore, only two serious rivals to *d*-tubocurarine chloride have emerged from the natural alkaloids and their derivatives—the methyl ether of *d*-T.C. itself, and the β-erythroidines. Apart from questions of potency the compounds do not differ significantly from curare, and do not, for instance, eliminate the need for careful control of respiration. Information is not yet adequate to assess their side-actions in clinical use.

From the pharmacological side, *d*-T.C. is clearly not the ideal curarising agent, because of its ganglionic action and its power of liberating histamine. Its depression of the respiratory minute volume is probably less important, since modern methods of anæsthesia are fully adequate to maintaining artificial respiration without inconvenience. A

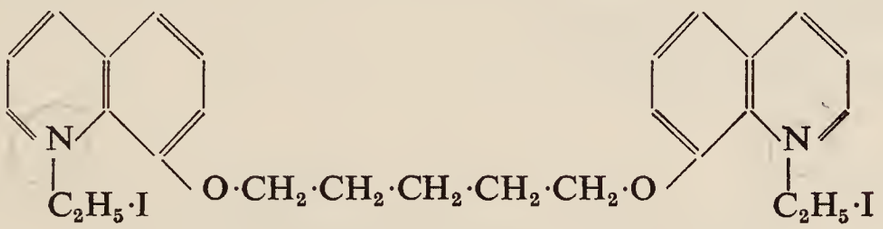
technical disadvantage of it is that some preparations cannot be given simultaneously with pentothal owing to mutual precipitation. Finally, it is a drug that is relatively expensive and difficult to prepare in the pure state.

Against all this, it is important to realise that it is with this compound that the use of curarising substances in anæsthesia has established itself; this rapid success, and the widespread search for substances of similar action are, indeed, sufficient testimony to its value.

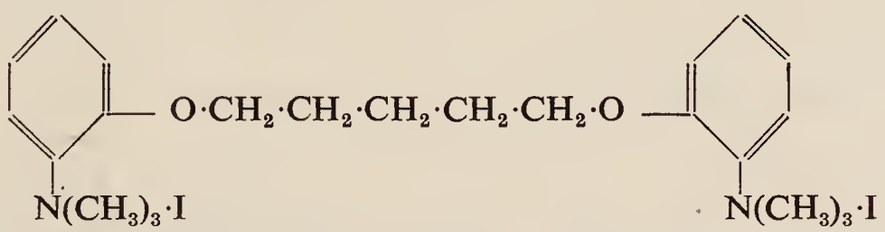
Synthetic Curarising Substances

1. Early Work. The first synthetic compound exhibiting curare-like activity, tetramethylammonium iodide, ($N(CH_3)_4.I.$) was made by Crum Brown and Fraser[2] in 1869, and provided them with a striking confirmation of their theories. Its curariform action is not strong and its power of stimulating autonomic ganglia is its most prominent action in the cat, followed by a weaker paralysing action on ganglia; it also possesses appreciable muscarine-like action (Burn and Dale[31]). Bacq and Brown[60] found that it could also elicit a contraction from mammalian striated muscle, and it is known to cause a contracture of frog's rectus. It is typical of the many related compounds that they possess, besides curariform activity, some or all of these other activities in some degree—some of them also possessing anticholinesterase potency. But none of the synthetic monoquaternary salts, whether simple tetralkylammonium halides or choline or betaine derivatives, proved to be sufficiently potent or free from side-actions even to approach the natural alkaloids.

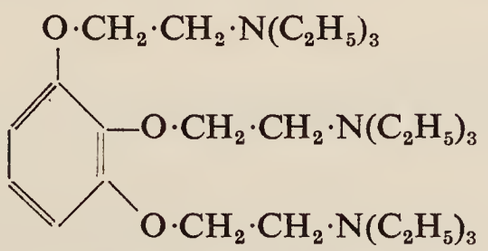
2. Bovet's Compounds. The work of Bovet and his colleagues marked an important advance. Taking as a model the structure of *d*-tubocurarine chloride (which had been almost completely defined by King at that time),



(a) 3381 R.P. Head drop dose = 0.25 mg./kg.



(b) 3565 R.P. Head drop dose = 0.2 mg./kg.



(c) 3697 R.P., 2559 F, flaxedil: Head drop dose = 0.5 mg./kg.

Fig. 2. Synthetic curarising compounds (Bovet).

simpler related structures were synthesised. Using this basic structure, variations of chain-length and quaternary substituents were studied. The first compound reported (Bovet, Courvoisier, Ducrot, and Horclois[9]), 3381 R.P., is also the first synthetic compound that resembles curare at all closely, in being potent and sensitive to anticholinesterases (Fig. 2(a)). It possesses, in addition, some anticholinesterase potency (Halpern, Benda and Bourdon[61]).

The next step was a further simplification of structure, as a result of which 3565 R.P. (see Fig. 2b) was described (Bovet, Courvoisier, Ducrot and Horclois[11]). This compound, too, is highly active, but does not possess anti-esterase action; it is antagonised by anti-esterases, and has some nicotinic action.

The third main series were the choline ethers of phenols and polyphenols (Bovet, Depierre and Lestrange[10]), of which the ethyl-choline triether of pyrogallol proved to be the most important (2559 F. or 3697 R.P., flaxedil. See Figure 2c). This possessed much the same actions as the other compounds with fewer side-actions.

Finally, an interesting series of choline esters has been built, also of considerable potency, but of transient action (Bovet, Bovet-Nitti, Guarino and Fusco[13]). Figure 3 gives a comparison of one of these with the corresponding ether and the directly substituted compound, together with their effective doses and duration of action.

	Head drop dose	*Duration of paralysis after 10 H. D. doses*
$COOCH_2 \cdot CH_2 \cdot N(C_2H_5)_3 \cdot I$	3 mg./kg.	10 minutes
$COOCH_2 \cdot CH_2 \cdot N(C_2H_5)_3 \cdot I$		
$O \cdot CH_2 \cdot CH_2 \cdot N(C_2H_5)_3 \cdot I$	3 mg./kg.	1 hour
$O \cdot CH_2 \cdot CH_2 \cdot N(C_2H_5)_3 \cdot I$		
$CH_2 \cdot CH_2 \cdot N(C_2H_5)_3 \cdot I$	3 mg./kg.	3 hours
$CH_2 \cdot CH_2 \cdot N(C_2H_5)_3 \cdot I$		

FIG. 3. Variation of duration of action with chemical structure.

Courvoisier and Ducrot[62] have shown that flaxedil (2559F) possesses little power to liberate histamine, although 3381 R.P. and 3365 R.P. are active in this respect. Depierre[33] has shown that it possesses little ganglionic action.

3. THE *bis*TRIMETHYLAMMONIUM SERIES. The most potent synthetic compound so far described is the decane derivative (C10) of an even simpler series, the *bis*trimethylammonium polymethylene salts. The curarising action of the series was reported independently by Barlow and Ing[63] and by Paton and Zaimis[64], and the latter authors have studied the pharmacological actions of the series in considerable detail (Paton and Zaimis[64,45]). Some of these actions of C10 have been already mentioned above, and will not be further discussed. But three points require further discussion.

(a) *Variation of potency with species and test object.* C10 is highly active in cat, 0·03 mg./kg. causing full neuromuscular paralysis; in rabbit 0·1 mg./kg. is required; in mouse 1 mg./kg., and in rat 5 mg./kg. In man a total dose of 3 mg. usually causes almost complete paralysis, and man thus corresponds very closely to the cat in sensitivity (Organe, Paton and Zaimis[65]); the equivalent dose of *d*-T.C. in a man is about 15 mg.

$$I{\cdot}N(CH_3)_3{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}CH_2{\cdot}N(CH_3)_3{\cdot}I$$

FIG. 4. Structure of *bis*Trimethylammonium decane diiodide (C10)

The comparison of *d*-T.C. with C10 further varies with the muscle used, and with the rate of stimulation. In the cat, using single twitches every 10 sec., C10 is 10-20 times as active as *d*-T.C. But if tetani are used this figure is at least halved, since *d*-T.C. depresses a tetanus much more than a twitch and C10 allows a tetanus to be fairly well sustained. In addition, the two compounds differ in their actions on different muscles; thus, in the cat, C10 paralyses tibialis more readily than soleus, while *d*-T.C. does the reverse. Finally, a test method such as that used by de Jalon[66], in which the antagonism of curare to acetylcholine on frog's rectus is employed, cannot be used at all, since C10 itself elicits a contracture and (by reason of its anti-esterase activity) actually potentiates acetylcholine. The findings on species difference are paralleled in other compounds. Collier, Paris and Woolf[52] have reported a species difference with the dimethyl ether of *d*-T.C., and Wien[67] has done so, using the isolated diaphragm, for some of Bovet's compounds. From such results, it is obvious that investigations of new compounds should be made by several test methods, and that in an assay the method used should be fully sensitive to all the substances likely to occur in the material under test. The fact that man closely resembles the cat in his sensitivity to C10 is an important argument for the use of cats at an early stage in any investigation. The species difference, and the different action on different muscles are fascinating problems for which there is no explanation at present.

(b) *Effect on respiration.* One of the remarkable features of the action of C10 in the cat is the failure to depress respiration significantly at a time when tibialis twitch is more than 95 per cent. paralysed (Paton and Zaimis[45]). This appears to be due to the fact that tetani are well-sustained, and that red muscles are less affected than white. The contrast with *d*-T.C. is sharp, for in the cat respiration may be depressed before any action on the twitch has appeared. But it is difficult to predict

the same sparing of respiration in relation to other muscular activities (e.g., abdominal relaxation in anæsthesia or the softening of therapeutic convulsions), in the absence of information about the physiological characteristics of the muscles involved and of the rate at which their motor units are excited.

(c) *Side actions.* Curarising compounds may depress the blood pressure by liberating histamine, by paralysing ganglia or by exerting a muscarine-like action. Curarising doses of C10 have no effect on the blood pressure of the anæsthetised cat, and large doses (at least 100 times the effective curarising dose) must be given to show such an action. It is at least 5 times less active in releasing histamine or causing ganglionic depression than the same weight of *d*-T.C., and its muscarine action is negligible. C10 has some anticholinesterase action, but this does not appear to cause any undesirable effect. In man, there is no interference with sensation or consciousness after an intravenous dose of C10 sufficient to cause almost complete paralysis (Organe, Paton and Zaimis[65]).

Discussion

There is much that is confusing in recent developments, which cannot be discussed here. But one feature may be stressed; this is the remarkable diversity of actions among the various compounds studied. Their variation in activity on different species, on different muscles, and on different synapses; their differences in side-actions, and their wide disparity in chemical structure:—all these are hopeful prognostics of yet other compounds with useful selective actions Before these are likely to be discovered, however, fundamental work must be done on the reasons for these diversities of action, about which we know little, and such fundamental research is the most urgent need. There are, further, many more practical requirements to be satisfied, such as a satisfactory method of prolonging the action of these drugs; compounds active by mouth and safe to use; or an antagonist to the activity of curarising agents which is free of side-actions.

To the pharmacologist, however, the most absorbing question remains that of the relation of the structure of these compounds to their pharmacological action. No field has proved more hazardous than that of the onium salts, in which to venture generalisations. Many of the important anomalies are reviewed by Ing[14]. Certain broad statements, however, can be made.

(1) Crum Brown and Fraser's generalisation of 1868 still remains remarkably true. The only important exception to it is that of the properties of the *Erythrina* alkaloids; and in ignorance of their structure it is impossible to say how much of an exception they represent. There is little doubt that the most promising approach, in devising substances of curarising potency, is still by way of quaternary salts; and among these, there is still no rival to the salts of quaternary nitrogen.

(2) The introduction of a second or even a third quaternary group into the molecule appears to be an important element in producing compounds of high curarising potency: all the compounds of known

chemical structure active in a dose of 1 mg./kg. or less have two or more such groups. Possible reasons for this enhancement of potency are not far to seek in the favourable effects such additions have on the attachment of the drugs to a receptor surface.

(3) The introduction of further quaternary groups has another effect: the monoquaternary compounds active on the neuromuscular junction are commonly like tetramethylammonium iodide, particularly in possessing stimulant nicotine-like and muscarine-like actions. In *bis*-quaternary salts these actions are replaced by weak anti-esterase activity, ganglionic depression, and histamine liberation. These latter actions would, indeed, be a serious disadvantage; but *bis*quaternary or *ter*quaternary compounds have been obtained in which they are slight, although the compounds still possess high potency (C10 and flaxedil).

(4) A further significant point appears to be the spatial separation of the two quaternary groups. It can hardly be coincidental that in such active compounds as *d*-tubocurarine chloride and its methyl ether, Bovet's compounds 3381 R.P. and 3565 R.P., and C10, the quaternary groups should be separated by 10 to 11 atoms, particularly since shortening the chain in the *bis*trimethylammonium series to less than 7 carbon atoms almost completely abolishes activity. The implications of this require further study, but the suggestion certainly appears that the ability to interfere with neuromuscular transmission depends not only on certain characteristic polar groups but also on their characteristic spatial location. It is certain that other considerations than distance are also concerned; thus, King[4] reports that *l*-tubocurarine chloride is 30 to 60 times weaker than the dextro-rotatory isomer.

Finally, it is worth pointing out that the diversity of structure exhibiting curarising activity suggests that the specific characteristics of such compounds differ in some important way from those of (for instance) muscarine-like substances. Pfeiffer[68] has drawn some striking analogies between the distances separating certain prosthetic groups (the nitrogen atom and two oxygen atoms) in muscarine-like and atropine-like substances, and the distances between the same groups in acetylcholine. But no such analogy is at present visible among curarising substances. It is, indeed, possible that their specific activity is not dependent solely on a relationship to acetylcholine, but that resemblance to some other physiologically active cation must be considered. The papers of Ing and Wright still represent the most critical approach to the subject; recent attempts to apply modern concepts of atomic structure to the problem (Holmes, Jenden, Taylor[70]) do not advance much beyond the position of Ing and Wright.

References

1. Crum Brown and Fraser, *Trans. roy. Soc. Edinb.*, 1868, **25,** 151, 693.
2. Crum Brown and Fraser, *Proc. roy. Soc. Edinb.*, 1869, **6,** 556.
3. King, *J. chem. Soc.*, 1935, 1381.
4. King, *ibid.*, 1947, 936.
5. King, *ibid.*, 1948, 265.
6. Bennett, *Amer. J. Psychiat.*, 1941, **97,** 1040.
7. Griffith and Johnson, *Anesthesiology,* 1942, **3,** 418.

8. Wintersteiner and Dutcher, *Science,* 1943, **97,** 467.
9. Bovet, Courvoiser, Ducrot and Horclois, *C.R. Acad. Sci., Paris,* 1946, **223,** 597.
10. Bovet, Depierre and de Lestrange, *C.R. Acad. Sci., Paris,* 1947, **225,** 74.
11. Bovet, Courvoiser, Ducrot and Horclois, *C.R. Acad. Sci., Paris,* 1947, **224,** 1733.
12. Bovet and Bovet-Nitti, *Experientia,* 1948, **4,** 325.
13. Bovet, Bovet-Nitti, Guarino and Fusco, *Helv. Physiol. Acta,* 1948, **6,** C52.
14. Ing, *Physiol. Rev.,* 1936, **16,** 527.
15. MacIntyre, *Curare, Chicago,* 1947.
16. Craig, *Chem. Rev.,* 1948, **42,** 285.
17. Acheson, Harvey, Kuffler and Welsh, *Fed. Proc.,* 1948, **7,** 447, 458, 437, 435.
18. Dale, *Harvey Lect.,* 1936-1937, **32,** 229.
19. Feldberg, *Physiol. Rev.,* 1945, **25,** 596.
20. Brown, Paton, Vianna Dias, *J. Physiol.,* 1949 (in the press).
21. Brown, Dale and Feldberg, *J. Physiol.,* 1936, **87,** 394.
22. Riker and Wescoe, *J. Pharmacol.,* 1946, **88,** 58.
23. Harvey, *Bull. Johns Hopk. Hosp.,* 1939, **65,** 223.
24. Burgen, Dickens and Zattman, *J. Physiol.,* 1948, **107,** 41P.
25. Brown and Harvey, *J. Physiol.,* 1940, **97,** 330.
26. Brown and Vianna Dias, *Ann. Acad. Bras. Ciencias,* 1947, **19,** 359.
27. Brown and Feldberg, *J. Physiol.,* 1936, **86,** 10P.
28. Brown and Feldberg, *J. Physiol.,* 1936, **88,** 265.
29. Harvey and MacIntosh, *J. Physiol.,* 1940, **97,** 408.
30. Brown and Feldberg, *J. Physiol.,* 1936, **86,** 290.
31. Burn and Dale, *J. Pharmacol.,* 1915, **6,** 417.
32. Acheson and Moe, *J. Pharmacol.,* 1946, **87,** 220.
33. Depierre, *C.R. Acad. Sci., Paris,* 1947, **225,** 956.
34. Krogh, *Proc. roy. Soc.,* 1946, **133B,** 140.
35. Salama, *J. Physiol.,* 1948, **108,** 24P; *J. Physiol.,* 1949, **108** (in the press).
36. Alam, Anrep, Barsoum, Talaat and Weininger, *J. Physiol.,* 1939, **95,** 148.
37. Gregory and Schild, *Proc. XVII Internat. Physiol. Cong.,* 1947, 288.
38. Grob, Lilienthal and Harvey, *Bull. Johns Hopk. Hosp.,* 1947, **80,** 299.
39. Landmesser, *Anesthesiology,* 1947, **8,** 506.
40. Everett, *J. Pharmacol.,* 1948, **92,** 936.
41. Whitacre and Fisher, *Anesthesiology,* 1945, **6,** 124.
42. Prescott, Organe and Rowbotham, *Lancet,* 1946, **251,** 80.
43. Smith, Brown, Toman and Goodman, *Anesthesiology,* 1947, **8,** 1.
44. Kellgren, McGowan and Wood, *Brit. med. J.,* 1946, **ii,** 898.
45. Paton and Zaimis, *J. Physiol.,* 1949, **108,** 34P; *J. Physiol.,* 1949 (in the press).
46. Feldberg and Lin, *J. Physiol.,* 1948, **107,** 37P.
47. Gross and Cullen, *Anesthesiology,* 1945, **6,** 231.
48. Heymans, *Experientia,* 1946, **2,** 453.
49. Heymans, *Arch. neerl. Physiol.,* 1948, **28,** 419.
50. Kensler, *J. Pharmacol.,* 1949, **95,** 28.
51. Harris and Harris, *Proc. Soc. exp. Biol., N.Y.,* 1944, **56,** 223.
52. Collier, Paris and Woolf, *Nature,* 1948, **161,** 817.
53. Stoelting, Graf and Viera, *Proc. Soc. exp. Biol., N.Y.,* 1948, **69,** 565.
54. Harvey and Masland, *J. Pharmacol.,* 1941, **73,** 304.
55. Dripps and Sergent, *Anesthesiology,* 1947, **8,** 241.
56. Harvey, *Johns Hopkins Hosp. Bull.,* 1940, **66,** 52.
57. Marsh and Pelletier, *J. Pharmacol.,* 1948, **92,** 454.
58. Marsh, Sleeth and Tucker, *J. Pharmacol.,* 1948, **93,** 109.
59. Marsh, Herring and Sleeth, *J. Pharmacol.,* 1949, **95,** 100.
60. Bacq and Brown, *J. Physiol.,* 1937, **89,** 45.
61. Halpern, Benda and Bourdon, *C.R. Soc. Biol., Paris,* 1947, **141,** 979.
62. Courvoisier and Ducrot, *C.R. Soc. Biol., Paris,* 1942, **142,** 1209.
63. Barlow and Ing, *Nature,* 1948, **161,** 718; Barlow and Ing, *J. Pharmacol.,* 1948, **3,** 298.
64. Paton and Zaimis, *Nature,* 1948, **161,** 718; *Nature,* 1948, **162,** 810.
65. Organe, Paton and Zaimis, *Lancet,* 1949, **256,** 21.
66. de Jalon, *Quart. J. Pharm. Pharmacol.,* 1947, **20,** 28.
67. Wien, *J. Physiol.,* 1948, **107,** 44P.
68. Pfeiffer, *Science,* 1948, **107,** 94.
69. Ing and Wright, *Proc. roy. Soc.,* 1931, **109B,** 337; Ing and Wright, *Proc. roy. Soc.,* 1933, **114B,** 48.
70. Holmes, Jenden and Taylor, *Nature,* 1947, **159,** 86.

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